

WHAT IS CLAIMED IS:

- ✓ 1. A method of screening a chemical for its ability to enhance binding of a co-regulatory protein to a nuclear receptor or to a nuclear receptor ligand binding domain, wherein said method comprises:
- (a) cotransfected cells with i) a gene which expresses a co-regulatory protein comprising SDPPSPS (SEQ ID NO:5) and ii) a nucleic acid comprising a gene encoding said nuclear receptor or said nuclear receptor ligand binding domain to produce cotransfected cells which synthesize said co-regulatory protein and said nuclear receptor or said nuclear receptor ligand binding domain, and further wherein said cotransfected cells comprise a reporter gene the expression of which depends upon said co-regulatory protein binding to said nuclear receptor or to said nuclear receptor ligand binding domain;
- (b) growing a first portion of said cotransfected cells in the presence of said chemical;
- (c) growing a second portion of said cotransfected cells in the absence of said chemical; and
- (d) determining the level of expression of said reporter gene in each portion of cells; wherein if the level of expression of said reporter gene in said first portion of said cells is greater than the level of expression in said second portion of said cells then said chemical enhances binding of said co-regulatory protein to said nuclear receptor or to said nuclear receptor ligand binding domain.
2. The method of claim 1 wherein said co-regulatory protein is 35 kDa.
3. The method of claim 1 wherein said co-regulatory protein comprises SEQ ID NO:9.
4. The method of claim 1 wherein said co-regulatory protein consists of SEQ ID NO:8.
5. The method of claim 1 wherein said nuclear receptor is SF1 or ERR α 1.
6. The method of claim 1 wherein said nuclear receptor ligand binding domain is selected from the group consisting of ER, AR, GR, PR, TR, RAR and RXR.

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7. The method of claim 1 wherein said nuclear receptor comprises a binding domain selected from the group consisting of TR, PR and RXR.
8. The method of claim 1 wherein expression of said reporter gene causes production of histidine.
9. The method of claim 1 wherein said reporter gene is CAT.
10. The method of claim 1 wherein said cells are yeast cells or human cells.
11. The method of claim 1 wherein said nuclear receptor is capable of binding to an aromatase gene.
12. A method of screening a chemical for its ability to inhibit binding of a co-regulatory protein to a nuclear receptor or to a nuclear receptor ligand binding domain, wherein said method comprises:
- (a) cotransfected cells with i) a gene which expresses a co-regulatory protein comprising SDPPSPS (SEQ ID NO:5) and ii) a nucleic acid comprising a gene encoding said nuclear receptor or said nuclear receptor ligand binding domain to produce cotransfected cells which synthesize said co-regulatory protein and said nuclear receptor or said nuclear receptor ligand binding domain, and further wherein said cotransfected cells comprise a reporter gene the expression of which depends upon said co-regulatory protein binding to said nuclear receptor or to said nuclear receptor ligand binding domain;
- (b) growing a first portion of said cotransfected cells in the presence of said chemical;
- (c) growing a second portion of said cotransfected cells in the absence of said chemical; and
- (d) determining the level of expression of said reporter gene in each portion of said cells, wherein if the level of expression of said reporter gene in said first portion of said cells is less than the level of expression in said second portion of said cells then said chemical inhibits binding of said co-regulatory protein to said nuclear receptor or to said nuclear receptor ligand binding domain.

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13. The method of claim 12 wherein said co-regulatory protein is 35 kDa.
14. The method of claim 12 wherein said co-regulatory protein comprises SEQ ID NO:9.
15. The method of claim 12 wherein said co-regulatory protein consists of SEQ ID NO:8.
16. The method of claim 12 wherein said nuclear receptor is SF1 or ERR α 1.
17. The method of claim 12 wherein said nuclear receptor ligand binding domain is selected from the group consisting of ER, AR, GR, PR, TR, RAR and RXR.
18. The method of claim 12 wherein said nuclear receptor comprises a binding domain selected from the group consisting of TR, PR and RXR.
19. The method of claim 12 wherein expression of said reporter gene causes production of histidine.
20. The method of claim 12 wherein said reporter gene is CAT.
21. The method of claim 12 wherein said cells are yeast cells or human cells.
22. The method of claim 12 wherein said nuclear receptor is capable of binding to an aromatase gene.
23. The method of claim 12 wherein a ligand which enhances binding of said co-regulatory protein to said nuclear receptor or to said nuclear receptor ligand binding domain is present.
24. The method of claim 23 wherein said ligand is estradiol, deoxycorticosterone, progesterone, dihydrotestosterone, T3, all-*trans*-retinoic acid or 9-*cis*-retinoic acid.

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- ✓ 25. A method of screening a test chemical to determine if it has activity similar to a known chemical, wherein said method comprises:
- (a) cotransfected cells with i) a gene which expresses a co-regulatory protein comprising SDPPSPS (SEQ ID NO:5) and ii) a nucleic acid comprising a gene encoding a nuclear receptor or said nuclear receptor ligand binding domain to produce cotransfected cells which synthesize said co-regulatory protein and said nuclear receptor or said nuclear receptor ligand binding domain, and further wherein said cotransfected cells comprise a reporter gene the expression of which depends upon said co-regulatory protein binding to said nuclear receptor or to said nuclear receptor ligand binding domain, and further wherein said co-regulatory protein binds to said nuclear receptor or to said nuclear receptor ligand binding domain in the presence of said known chemical;
 - (b) growing a first portion of said cotransfected cells in the presence of said test chemical;
 - (c) growing a second portion of said cotransfected cells in the absence of said chemical; and
 - (d) determining the level of expression of said reporter gene in each portion of cells; wherein if the level of expression of said reporter gene in said first portion of said cells is greater than the level of expression in said second portion of said cells then said test chemical has activity similar to said known chemical.
26. The method of claim 25 wherein said co-regulatory protein is 35 kDa.
27. The method of claim 25 wherein said co-regulatory protein comprises SEQ ID NO:9.
28. The method of claim 25 wherein said co-regulatory protein consists of SEQ ID NO:8.
29. The method of claim 25 wherein said nuclear receptor ligand binding domain and said known chemical are selected from the sets of (i) ER and estradiol, (ii) GR and deoxycorticosterone, (iii) AR and dihydrotestosterone, (iv) PR and progesterone, (v) TR and T3, (vi) RAR and all-*trans*-retinoic acid, and (vii) RXR and 9-*cis*-retinoic acid.

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30. The method of claim 25 wherein expression of said reporter gene causes production of histidine.

31. The method of claim 25 wherein said reporter gene is CAT.

32. The method of claim 25 wherein said cells are yeast cells or human cells.

33. The method of claim 25 wherein said nuclear receptor is capable of binding to an aromatase gene.

34. A method of determining a concentration of a ligand or a hormone in a tissue sample, wherein said method comprises:

(a) cotransfected cells with i) a gene which expresses a co-regulatory protein comprising SDPPSPS (SEQ ID NO:5) and ii) a nucleic acid comprising a gene encoding a nuclear receptor or said nuclear receptor ligand binding domain to produce cotransfected cells which synthesize said co-regulatory protein and said nuclear receptor or said nuclear receptor ligand binding domain, and further wherein said cotransfected cells comprise a reporter gene the expression of which depends upon said co-regulatory protein binding to said nuclear receptor or to said nuclear receptor ligand binding domain, and further wherein said co-regulatory protein binds to said nuclear receptor or to said nuclear receptor ligand binding domain in the presence of said ligand or hormone;

(b) preparing an extract of said tissue sample;

(c) growing a first portion of said cotransfected cells in the presence of said tissue extract;

(d) growing second portions of said cotransfected cells in the presence of known concentrations of said ligand or hormone; and

(e) determining the level of expression of said reporter gene in each portion of cells; wherein the concentration of said ligand or said hormone in said tissue extract can be determined by comparison of the expression level of said reporter gene in said first portion with the expression levels of said reporter gene in said second portions.

35. The method of claim 34 wherein said co-regulatory protein is 35 kDa.

36. The method of claim 34 wherein said co-regulatory protein comprises SEQ ID NO:9.
37. The method of claim 34 wherein said co-regulatory protein consists of SEQ ID NO:8.
38. The method of claim 34 wherein said nuclear receptor ligand binding domain and said ligand or hormone are selected from the sets of (i) ER and estradiol, (ii) GR and deoxycorticosterone, (iii) AR and dihydrotestosterone, (iv) PR and progesterone, (v) TR and T3, (vi) RAR and all-*trans*-retinoic acid, and (vii) RXR and 9-*cis*-retinoic acid.
39. The method of claim 34 wherein expression of said reporter gene causes production of histidine.
40. The method of claim 34 wherein said reporter gene is CAT.
41. The method of claim 34 wherein said cells are yeast cells or human cells.
42. The method of claim 34 wherein said nuclear receptor is capable of binding to an aromatase gene.
43. A method of screening for a protein which interacts with a chemical, wherein said method comprises:
(a) cotransfected cells with i) a gene which expresses a co-regulatory protein comprising SDPPSPS (SEQ ID NO:5) and ii) a library of nucleic acids to produce a library of cotransfected cells which synthesize said co-regulatory protein and said library of nucleic acids, and further wherein said cotransfected cells comprise a reporter gene the expression of which depends upon said co-regulatory protein binding to a nuclear receptor or to a nuclear receptor ligand binding domain, and further wherein said co-regulatory protein binds to said nuclear receptor or to said nuclear receptor ligand binding domain in the presence of said chemical;
(b) growing a first portion of said cotransfected cells in the absence of said chemical;

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(c) growing a second portion of said cotransfected cells in the presence of said chemical, wherein said second portion is a replicate of said first portion; and
 (d) comparing the level of expression of said reporter gene in individual colonies of said first portion and in said second portion; wherein if a colony of said second portion expresses said reporter gene at a higher level than its corresponding replicate colony in said first portion then said colony comprises a gene from said library encoding a protein which interacts with said chemical.

- Part B1*
44. The method of claim 43 wherein said co-regulatory protein is 35 kDa.
45. The method of claim 43 wherein said co-regulatory protein comprises SEQ ID NO:9.
46. The method of claim 43 wherein said co-regulatory protein consists of SEQ ID NO:8.
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47. The method of claim 43 wherein said nuclear receptor ligand binding domain and said ligand or hormone are selected from the sets of (i) ER and estradiol, (ii) GR and deoxycorticosterone, (iii) AR and dihydrotestosterone, (iv) PR and progesterone, (v) TR and T3, (vi) RAR and all-trans-retinoic acid, and (vii) RXR and 9-cis-retinoic acid.
- Part B2*
48. The method of claim 43 wherein expression of said reporter gene causes production of histidine.
49. The method of claim 43 wherein said reporter gene is CAT.
50. The method of claim 43 wherein said cells are yeast cells or human cells.
51. The method of claim 43 wherein said nuclear receptor is capable of binding to an aromatase gene.
- Add B3*

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- ✓ 52. A method of screening for a protein which interacts with a chemical, wherein said method comprises:
- (a) cotransfected cells with i) a gene which expresses a co-regulatory protein comprising SDPPSPS (SEQ ID NO:5) and ii) a library of nucleic acids to produce a library of cotransfected cells which synthesize said co-regulatory protein and said library of nucleic acids, and further wherein said cotransfected cells comprise a reporter gene the expression of which depends upon said co-regulatory protein binding to a nuclear receptor or to a nuclear receptor ligand binding domain, and further wherein said co-regulatory protein binds to said nuclear receptor or to said nuclear receptor ligand binding domain in the presence of said chemical;
- (b) growing colonies of said cotransfected cells in the presence of said chemical; and
- (c) determining the level of expression of said reporter gene in individual colonies of cells;
- wherein a colony of cells which expresses said reporter gene comprises a gene from said library encoding a protein which interacts with said chemical.

Add E1

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